Amendment and Response

Applicants: Adler et al. Serial No.: 10/091,166 Filed: March 5, 2002

For: NOVEL BETA-DEFENSINS

In the Specification

The paragraphs shown below are amended to indicate changes made. Amendments to the following are indicated by underlining what has been added and striking-through what has been deleted.

Please replace the paragraph beginning at page 26 line 14, with the following rewritten paragraph.

The results showed that the zamp1 gene maps 33.5 cR_3000 from the top of the human chromosome 8 linkage group on the WICGR radiation hybrid map. Proximal and distal framework markers were CHLC.GATA62D10 and WI-3823 (D8S1511), respectively. The use of surrounding markers positions the zamp1 gene in the 8p23.3-p23.2 region on the integrated LDB chromosome 8 map (located on the Internet, e.g., a public server of The Genetic Location Database, University of Southhampton, WWW server: http://cedar.genetics.soton.ac.uk/public_html/).

Please replace the paragraph beginning at page 36 line 16, with the following rewritten paragraph.

As an illustration, a nucleic acid molecule encoding a variant zamp1 polypeptide can be hybridized with a nucleic acid molecule having at least a portion of the nucleotide sequence of SEQ ID NOs:1, 4, 9 or 11 (or their complements) at 42°C overnight in a solution comprising 50% formamide, 5x SSC (1x SSC: 0.15 M sodium chloride and 15 mM sodium citrate), 50 mM sodium phosphate (pH 7.6), 5x Denhardt's solution (100x Denhardt's solution: 2% (w/v) FicoHFICOLL® 400, 2% (w/v) polyvinylpyrrolidone, and 2% (w/v) bovine serum albumin), 10% dextran sulfate, and 20 µg/ml denatured, sheared salmon sperm DNA. One of skill in the art can devise variations of these hybridization conditions. For example, the hybridization mixture can be incubated at a higher or lower temperature, such as about 65°C, in a solution that does not contain formamide. Moreover, premixed hybridization solutions are available (e.g., EXPRESSHYB Hybridization Solution from CLONTECH Laboratories, Inc.), and hybridization can be performed according to the manufacturer's instructions.

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Please replace the paragraph beginning at page 58 line 34, with the following rewritten paragraph.

Expressed recombinant zamp1 polypeptides (or chimeric zamp1 polypeptides) can be purified using fractionation and/or conventional purification methods and media. Ammonium sulfate precipitation and acid or chaotrope extraction may be used for fractionation of samples. Exemplary purification steps may include hydroxyapatite, size exclusion, FPLC and reverse-phase high performance liquid chromatography. Suitable anion exchange media include derivatized dextrans, agarose, cellulose, polyacrylamide, specialty silicas, and the like. PEI, DEAE, QAE and Q derivatives are preferred, with DEAE Fast-Flow SepharoseSEPHAROSE® (Pharmacia, Piscataway, NJ) being particularly preferred. Exemplary chromatographic media include those media derivatized with phenyl, butyl, or Phenyl——SepharoseSEPHAROSE® FF (Pharmacia), octyl groups, such Toyopearl TOYOPEARL® butyl 650 (Toso Haas, Montgomeryville, PA), Octyl-SepharoseSEPHAROSE® (Pharmacia) and the like; or polyacrylic resins, such as Amberchrom CG 71 (Toso Haas) and the like. Suitable solid supports include glass beads, silica-based resins, cellulosic resins, agarose beads, cross-linked agarose beads, polystyrene beads, cross-linked polyacrylamide resins and the like that are insoluble under the conditions in which they are to be used. These supports may be modified with reactive groups that allow attachment of proteins by amino groups, carboxyl groups, sulfhydryl groups, hydroxyl groups and/or carbohydrate moieties. Examples of coupling chemistries include cyanogen bromide activation, N-hydroxysuccinimide activation, epoxide activation, sulfhydryl activation, hydrazide activation, and carboxyl and amino derivatives for carbodiimide coupling chemistries. These and other solid media are well known and widely used in the art, and are available from commercial suppliers. Methods for binding receptor polypeptides to support media are well known in the art. Selection of a particular method is a matter of routine design and is determined in part by the properties of the chosen support. See, for example, Affinity Chromatography: Principles & Methods, Pharmacia LKB Biotechnology, Uppsala, Sweden, 1988.

Please replace the paragraph beginning at page 72 line 30, with the following rewritten paragraph.

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The zamp1 gene was mapped to chromosome 8 using the commercially available "GeneBridge 4 Radiation Hybrid Panel" (Research Genetics, Inc., Huntsville, AL). The GeneBridge 4 Radiation Hybrid Panel contains PCRable DNAs from each of 93 radiation hybrid clones, plus two control DNAs (the HFL donor and the A23 recipient). A publicly available WWW server (http://www-genome.wi.mit.edu/egi-bin/contig/rhmapper.pl) located on the Internet allows mapping relative to the Whitehead Institute/MIT Center for Genome Research's radiation hybrid map of the human genome (the "WICGR" radiation hybrid map) which was constructed with the GeneBridge 4 Radiation Hybrid Panel.

Please replace the paragraph beginning at page 73 line 26, with the following rewritten paragraph.

The results showed that the zamp1 gene maps 33.5 cR_3000 from the top of the human chromosome 8 linkage group on the WICGR radiation hybrid map. Proximal and distal framework markers were CHLC.GATA62D10 and WI-3823 (D8S1511), respectively. The use of surrounding markers positions the zamp1 gene in the 8p23.3-p23.2 region on the integrated LDB chromosome 8 map (The Genetic Location Database, University of Southhampton, WWW server: http://cedar.genetics.soton.ac.uk/ public_html/located on the Internet).